	Type	L #	Hits	Search Text	DBs	Time Stamp	Comments	Error Definition	Errors
1	BRS	L1	0	nanomag	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:03			0
2	BRS	L2	301661	protein or proteinaceous	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:04			0
3	BRS	L3	1047339	isolat$3 or purif$7	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:05			0
4	BRS	L4	84557	2 same 3	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:05			0
5	BRS	L5	22692	magnetic adj particle	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:06			0
6	BRS	L6	4693	magnetic adj bead	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:08			0
7	BRS	L7	573	4 same (5 or 6)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:09			0
8	BRS	L8	49345	hydrophobic same hydrophilic	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:09			0
9	BRS	L9	10	7 same 8	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:33			0

	Type	L #	Hits	Search Text	DBs	Time Stamp	Comments	Error Definition	Errors
10	BRS	L10	1053	agarose same hydrophobic	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:33			0
11	BRS	L11	11108	silica same (magnetic or magnetite)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:34			0
12	BRS	L12	2708	(silica adj gel) same (reverse adj phase)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:35			0
13	BRS	L13	0	7 same (10 or 12)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:36			0
14	BRS	L14	30	4 same 11	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:36			0
15	BRS	L15	313	(C-18 or C-8) same ((silica adj gel) same (reverse adj phase))	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:53			0
16	BRS	L16	0	7 same 15	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:54			0
17	BRS	L17	20759	mass adj spectrometry	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:55			0
18	BRS	L18	8	(7 or 9 or 14) same 17	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:56			0

	Type	L #	Hits	Search Text	DBs	Time Stamp	Comments	Error Definition	Errors
19	BRS	L19	3	rauth adj holger.in.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:58			0
20	BRS	L21	7	reinhardt adj richard.in.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 12:59			0
21	BRS	L22	6	nordhoff adj eckhard.in.	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 13:00			0
22	BRS	L23	1	(19 or 21 or 22) and (7 or 9 or 14 or 18)	USPAT; US-PGPUB; EPO; JPO; DERWENT	2003/02/24 13:01			0

=> d his

(FILE 'HOME' ENTERED AT 13:48:38 ON 24 FEB 2003)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE, SCISEARCH, AGRICOLA' ENTERED AT
13:49:06 ON 24 FEB 2003
L1 1 S NANOMAG
L2 7884617 S PROTEIN OR PROTEINACEOUS OR PEPTIDE
L4 5036901 S ISOLAT? OR PURIF?
L5 1223596 S L2 (P) L4
L6 15086 S (MAGNETIC PARTICLE) OR (MAGNETIC BEAD)
L7 847 S L5 (P) L6
L8 1856 S AGAROSE (P) HYDROPHOBIC
L9 937 S (SILICA GEL) (P) (REVERSE PHASE)
L10 0 S L7 (P) (L8 OR L9)
L11 231 S SILICA (P) MAGNETIC (P) (HYDROPHOBIC)
L12 0 S L5 (P) L11
L13 371943 S HYDROPHOBIC OR HYDROPHILIC
L14 7 S L7 (P) L13
L15 4 DUPLICATE REMOVE L14 (3 DUPLICATES REMOVED)
L16 60864 S MASS SPECTROSCOPY
L17 2 S L7 (P) L16
L18 2 DUPLICATE REMOVE L17 (0 DUPLICATES REMOVED)
L19 2 S L17 NOT L15

=> log y

```
FILE 'HOME' ENTERED AT 13:48:38 ON 24 FEB 2003

=> file medline caplus biosis embase scisearch agricola
COST IN U.S. DOLLARS                                SINCE FILE      TOTAL
                                                       ENTRY    SESSION
FULL ESTIMATED COST                                     0.21       0.21

FILE 'MEDLINE' ENTERED AT 13:49:06 ON 24 FEB 2003

FILE 'CAPLUS' ENTERED AT 13:49:06 ON 24 FEB 2003
USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
COPYRIGHT (C) 2003 AMERICAN CHEMICAL SOCIETY (ACS)

FILE 'BIOSIS' ENTERED AT 13:49:06 ON 24 FEB 2003
COPYRIGHT (C) 2003 BIOLOGICAL ABSTRACTS INC.(R)

FILE 'EMBASE' ENTERED AT 13:49:06 ON 24 FEB 2003
COPYRIGHT (C) 2003 Elsevier Science B.V.  All rights reserved.

FILE 'SCISEARCH' ENTERED AT 13:49:06 ON 24 FEB 2003
COPYRIGHT (C) 2003 Institute for Scientific Information (ISI) (R)

FILE 'AGRICOLA' ENTERED AT 13:49:06 ON 24 FEB 2003

=> s nanomag
L1            1 NANOMAG

=> d l1 1 ibib abs

L1   ANSWER 1 OF 1  CAPLUS  COPYRIGHT 2003 ACS
ACCESSION NUMBER:         2001:535623  CAPLUS
DOCUMENT NUMBER:          135:315508
TITLE:                    High-Tc SQUID system for biological immunoassays
AUTHOR(S):                Enpuku, K.; Hotta, M.; Nakahodo, A.
CORPORATE SOURCE:         Department of Electronics, Kyushu University,
                          Higashi-ku, Fukuoka, 812-8581, Japan
SOURCE:                   Physica C: Superconductivity and Its Applications
                          (Amsterdam, Netherlands) (2001), 357-360(Pt. 2),
                          1462-1465
                          CODEN: PHYCE6; ISSN: 0921-4534
PUBLISHER:                Elsevier Science B.V.
DOCUMENT TYPE:            Journal
LANGUAGE:                 English
AB   A high-Tc SQUID system is developed for the application to biol.
     immunoassay, i.e., for the detection of an antigen with a magnetically
     labeled antibody using ferric oxide.  In order to improve the system
     performance, two issues were studied.  One is the use of a gradiometer to
     suppress the system noise due to residual environmental noise.  The noise
     of the gradiometer system is five times smaller than that of the
     magnetometer system.  The other is to increase the signal field from the
     magnetic marker.  The signal can be increased by increasing an applied
     field Bex or by using remanence of the marker.  If these improvements were
     fully developed, it will be possible to develop the system that is 100
     times more sensitive than the conventional optical method.
REFERENCE COUNT:          9     THERE ARE 9 CITED REFERENCES AVAILABLE FOR THIS
                                RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT


=> s protein or proteinaceous or peptide
   4 FILES SEARCHED...
L2      7884617 PROTEIN OR PROTEINACEOUS OR PEPTIDE

=> s siolat? or purif?
L3      2053372 SIOLAT? OR PURIF?

=> s isolat? or purif?
L4      5036901 ISOLAT? OR PURIF?

=> s l2 (p) l4
```

```
L5     1223596 L2 (P) L4

=> s (magnetic particle) or (magnetic bead)
L6       15086 (MAGNETIC PARTICLE) OR (MAGNETIC BEAD)

=> s l5 (p) l6
L7         847 L5 (P) L6

=> s agarose (p) hydrophobic
L8        1856 AGAROSE (P) HYDROPHOBIC

=> s (silica gel) (p) (reverse phase)
L9         937 (SILICA GEL) (P) (REVERSE PHASE)

=> s l7 (p) (l8 or l9)
L10          0 L7 (P) (L8 OR L9)

=> s silica (p) magnetic (p) (hydrophobic)
L11        231 SILICA (P) MAGNETIC (P) (HYDROPHOBIC)

=> s l5 (p) l11
L12          0 L5 (P) L11

=> d his

     (FILE 'HOME' ENTERED AT 13:48:38 ON 24 FEB 2003)

     FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE, SCISEARCH, AGRICOLA' ENTERED AT
     13:49:06 ON 24 FEB 2003
L1           1 S NANOMAG
L2     7884617 S PROTEIN OR PROTEINACEOUS OR PEPTIDE
L3     2053372 S SIOLAT? OR PURIF?
L4     5036901 S ISOLAT? OR PURIF?
L5     1223596 S L2 (P) L4
L6       15086 S (MAGNETIC PARTICLE) OR (MAGNETIC BEAD)
L7         847 S L5 (P) L6
L8        1856 S AGAROSE (P) HYDROPHOBIC
L9         937 S (SILICA GEL) (P) (REVERSE PHASE)
L10          0 S L7 (P) (L8 OR L9)
L11        231 S SILICA (P) MAGNETIC (P) (HYDROPHOBIC)
L12          0 S L5 (P) L11

=> s hydrophobic or hydrophilic
L13    371943 HYDROPHOBIC OR HYDROPHILIC

=> s l7 (p) l13
L14          7 L7 (P) L13

=> duplicate remove l14
DUPLICATE PREFERENCE IS 'MEDLINE, CAPLUS, BIOSIS, EMBASE, SCISEARCH'
KEEP DUPLICATES FROM MORE THAN ONE FILE?  Y/(N):n
PROCESSING COMPLETED FOR L14
L15          4 DUPLICATE REMOVE L14 (3 DUPLICATES REMOVED)

=> d l15 1-4 ibib abs

L15  ANSWER 1 OF 4  SCISEARCH  COPYRIGHT 2003 ISI (R)
ACCESSION NUMBER:    97:473161  SCISEARCH
THE GENUINE ARTICLE: XE529
TITLE:               Genetic analysis of biomagnetic crystal formation
AUTHOR:              Matsunaga T (Reprint); Tsujimura N; Kamiya S
CORPORATE SOURCE:    TOKYO UNIV AGR & TECHNOL, DEPT BIOTECHNOL, 2-24-16 NAKA
                     CHO, KOGANEI, TOKYO 184, JAPAN (Reprint); TDK AKITA LAB
                     CORP, NIKAHO, AKITA 01804, JAPAN
COUNTRY OF AUTHOR:   JAPAN
SOURCE:              JOURNAL DE PHYSIQUE IV, (MAR 1997) Vol. 7, No. C1, pp.
                     651-654.
                     Publisher: EDITIONS PHYSIQUE, Z I DE COURTABOEUF AVE 7 AV
                     DU HOGGAR, BP 112, 91944 LES ULIS CEDEX, FRANCE.
                     ISSN: 1155-4339.
DOCUMENT TYPE:       Article; Journal
FILE SEGMENT:        PHYS
```

LANGUAGE: English
REFERENCE COUNT: 16
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Magnetospirillum sp. AMB-1 is a freshwater magnetic bacterium which synthesizes intracellular particles of magnetite (Fe3O4). A genomic DNA fragment required for synthesis of ***magnetic*** ***particles*** was previously ***isolated*** from a non-magnetic transposon Tn5 mutant. We have determined the complete nucleotide sequence of this fragment. The 2975 bp region contains two putative open reading frames (ORFs). One ORF, designated magA, encodes a polypeptide which is homologous to the cation efflux ***proteins*** , the Escherichia coli potassium ion translocating ***protein*** , KefC, and the putative Na+/H+-antiporter, NapA, from Enterococcus hirae. Intracellular localization of the MagA ***protein*** was studied using magA - luc fusion ***proteins*** . The luc gene was cloned downstream of the magA ***hydrophilic*** C-terminal domain. The fusion ***protein*** was also detected on the surface of the lipid bilayer covering the ***magnetic*** ***particles*** . These results suggest that MagA is a membrane-bound ***protein*** . Vesicles which contained MagA ***protein*** exhibited iron accumulation ability. We consider that the MagA ***protein*** is an iron transporter involved in the synthesis of ***magnetic*** ***particles*** in AMB-1.

L15 ANSWER 2 OF 4 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1998:52870 CAPLUS
DOCUMENT NUMBER: 128:163278
TITLE: Production of a protein (enzyme, antibody, protein A)-magnetite complex by genetically engineered magnetic bacteria Magnetospirillum sp. AMB-1
AUTHOR(S): Matsunaga, Tadashi; Kamiya, Shinji; Tsujimura, Noriyuki
CORPORATE SOURCE: Dep. Biotechnology, Tokyo Univ. Agriculture and Technology, Tokyo, 184, Japan
SOURCE: Scientific and Clinical Applications of Magnetic Carriers, Proceedings of the International Conference on Scientific and Clinical Applications of Magnetic Carriers, 1st, Rostock, Germany, Sept. 5-7, 1996 (1997), Meeting Date 1996, 287-294. Editor(s): Haefeli, Urs. Plenum: New York, N. Y.
CODEN: 65MWAX
DOCUMENT TYPE: Conference; General Review
LANGUAGE: English

AB A review, with 15 refs. Magnetospirillum sp. AMB-1 is a magnetic bacterium which synthesizes interacellular particles of magnetite (Fe3O4). A genomic DNA fragment required for the synthesis of ***magnetic*** ***particles*** was previously ***isolated*** from this strain. The complete nucleotide sequence of this fragment has been detd. by us. An open reading frame (ORF), designated magA, encodes a polypeptide which represents an iron transport ***protein*** . Intracellular localization of the MagA ***protein*** was studied using magA-luc fusion ***proteins*** . The luc gene was cloned downstream of the magA ***hydrophilic*** C-terminal domain. The fusion ***protein*** was detected on the surface of the lipid bilayer covering the ***magnetic*** ***particles*** . The MagA ***protein*** on the bacterial ***magnetic*** ***particle*** (BMP) was detected by immunoassay using an anti-luciferase antibody. The N- and C-termini of MagA ***protein*** were found outside the BMP membrane. These results show the utility of magA fusions for detecting multifunctional ***proteins*** on BMP. Recombinant ***protein*** -BMP complex prodn. has been carried out using the fed-batch culture by adding nitric acid and succinate as nitrogen and carbon source. These results suggest that genetic engineered magnetic bacteria are useful for the prodn. of ***protein*** (enzyme, antibody, ***protein*** A)-magnetite complexes.

L15 ANSWER 3 OF 4 CAPLUS COPYRIGHT 2003 ACS
ACCESSION NUMBER: 1997:438575 CAPLUS
DOCUMENT NUMBER: 127:76858
TITLE: Genetic analysis of biomagnetic crystal formation
AUTHOR(S): Matsunaga, T.; Tsujimura, N.; Kamiya, S.
CORPORATE SOURCE: Department of Biotechnology, Tokyo University of Agriculture and Technology, Koganei, 184, Japan
SOURCE: Journal de Physique IV (1997), 7(C1, 7th International

Conference on Ferrites, 1996), C1/651-C1/654
CODEN: JPI[redacted] ISSN: 1155-4339
PUBLISHER: Editions de Physique
DOCUMENT TYPE: Journal
LANGUAGE: English
AB Magnetospirillum sp. AMB-1 is a freshwater magnetic bacterium which synthesizes intracellular particles of magnetite (Fe3O4). A genomic DNA fragment required for synthesis of ***magnetic*** ***particles*** was previously ***isolated*** from a non-magnetic transposon Tn5 mutant. We have detd. the complete nucleotide sequence of this fragment. The 2975 bp region contains two putative open reading frames (ORFs). One ORF, designated magA, encodes a polypeptide which is homologous to the cation efflux ***proteins*** , the Escherichia coli potassium ion translocating ***protein*** , KefC, and the putative Na+/H+-antiporter, NapA, from Enterococcus hirae. Intracellular localization of the MagA ***protein*** was studied using magA-luc fusion ***proteins*** . The luc gene was cloned downstream of the magA ***hydrophilic*** C-terminal domain. The fusion ***protein*** was also detected on the surface of the lipid bilayer covering the ***magnetic*** ***particles*** . These results suggest that MagA is a membrane-bound ***protein*** . Vesicles which contained MagA ***protein*** exhibited iron accumulation ability. We consider that the MagA ***protein*** is an iron transporter involved in the synthesis of ***magnetic*** ***particles*** in AMB-1.

L15 ANSWER 4 OF 4 MEDLINE DUPLICATE 1
ACCESSION NUMBER: 90277607 MEDLINE
DOCUMENT NUMBER: 90277607 PubMed ID: 2161829
TITLE: Apolipoprotein B is both integrated into and translocated across the endoplasmic reticulum membrane. Evidence for two functionally distinct pools.
AUTHOR: Davis R A; Thrift R N; Wu C C; Howell K E
CORPORATE SOURCE: Cell and Molecular Biology Unit, University of Colorado Medical School, Denver 80262.
CONTRACT NUMBER: DK34914 (NIDDK)
HL25596 (NHLBI)
HL41624 (NHLBI)
SOURCE: JOURNAL OF BIOLOGICAL CHEMISTRY, (1990 Jun 15) 265 (17) 10005-11.
Journal code: 2985121R. ISSN: 0021-9258.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199007
ENTRY DATE: Entered STN: 19900824
Last Updated on STN: 19970203
Entered Medline: 19900716
AB Apolipoprotein B (apoB), a ***protein*** containing several ***hydrophobic*** beta-sheet structures, is essential for the assembly of triglyceride-rich lipoproteins. Previously, we found that only a fraction of de novo synthesized apoB is secreted; the remainder is retained in the endoplasmic reticulum where it is degraded. To understand the basis for these observations, translocation, the first step in the secretory pathway, was examined. Translocation of apoB was determined by its sensitivity to degradation by the exogenous protease, trypsin. In rough microsomes, about half of the apoB was degraded by trypsin. In contrast, in Golgi fractions little (if any) apoB was accessible to trypsin. Essentially all of the apoB that was degraded was membrane bound. Monoclonal IgGs against either the N-terminal or C-terminal halves of apoB were bound to ***magnetic*** ***beads*** and used to immunoisolate microsomes. In contrast to the specific ability of the IgGs against apoB to ***isolate*** microsomes, little or no microsomes were ***isolated*** using nonimmune IgG and IgG against albumin. Since microsomes remained intact and oriented right-side out as demonstrated by the inability of trypsin both to degrade albumin and to affect the capacity of the intralumenal enzyme glucose-6-phosphatase to dephosphorylate mannose 6-phosphate, the data suggest that a pool of apoB is exposed on the cytoplasmic surface of the endoplasmic reticulum membrane. To determine if the trypsin-accessible pool of apoB is a transient form, pulse-chase experiments were performed. The results show that the percent of apoB that was trypsin accessible increased during the

first 20 min of the chase, suggesting that during this time the trypsin-accessible pool of apoB is not translocated (it does not become trypsin insensitive). Thus, in two in vivo models (cultured cells and rat liver) translocation of apoB is not quantitative. We propose that apoB translocation across the endoplasmic reticulum determines its entry into two functionally distinct pools. The intralumenal trypsin-insensitive pool participates in the assembly of very low density lipoprotein; the trypsin-accessible nontranslocated cytoplasmic pool is shunted into a degradative pathway. Regulated translocation of apoB may provide a unique mechanism with which to determine the rate of very low density lipoprotein assembly/secretion.

=> d his

(FILE 'HOME' ENTERED AT 13:48:38 ON 24 FEB 2003)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE, SCISEARCH, AGRICOLA' ENTERED AT 13:49:06 ON 24 FEB 2003

```
L1             1 S NANOMAG
L2       7884617 S PROTEIN OR PROTEINACEOUS OR PEPTIDE
L3       2053372 S SIOLAT? OR PURIF?
L4       5036901 S ISOLAT? OR PURIF?
L5       1223596 S L2 (P) L4
L6         15086 S (MAGNETIC PARTICLE) OR (MAGNETIC BEAD)
L7           847 S L5 (P) L6
L8          1856 S AGAROSE (P) HYDROPHOBIC
L9           937 S (SILICA GEL) (P) (REVERSE PHASE)
L10            0 S L7 (P) (L8 OR L9)
L11          231 S SILICA (P) MAGNETIC (P) (HYDROPHOBIC)
L12            0 S L5 (P) L11
L13       371943 S HYDROPHOBIC OR HYDROPHILIC
L14            7 S L7 (P) L13
L15            4 DUPLICATE REMOVE L14 (3 DUPLICATES REMOVED)

=> s mass spectroscopy
L16       60864 MASS SPECTROSCOPY

=> s l7 (p) l16
L17            2 L7 (P) L16

=> duplicate remove l17
PROCESSING COMPLETED FOR L17
L18            2 DUPLICATE REMOVE L17 (0 DUPLICATES REMOVED)

=> s l17 not l15
L19            2 L17 NOT L15

=> d l19 1-2 ibib abs

L19  ANSWER 1 OF 2  BIOSIS  COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER:   2002:597203  BIOSIS
DOCUMENT NUMBER:    PREV200200597203
TITLE:              Charging of a naturally occurring amber tRNA with lysine by
                    a dedicated aminoacyl-tRNA synthetase from Methanosarcina
                    barkeri.
AUTHOR(S):          James, C. M. (1); Srinivasan, G. (1); Krzycki, J. A. (1)
CORPORATE SOURCE:   (1) Ohio State University, Columbus, OH USA
SOURCE:             Abstracts of the General Meeting of the American Society
                    for Microbiology, (2002) Vol. 102, pp. 250.
                    http://www.asmusa.org/mtgsrc/generalmeeting.htm. print.
                    Meeting Info.: 102nd General Meeting of the American
                    Society for Microbiology Salt Lake City, UT, USA May 19-23,
                    2002 American Society for Microbiology
                    . ISSN: 1060-2011.
DOCUMENT TYPE:      Conference
LANGUAGE:           English
AB   Methanogenesis in Methanosarcina barkeri from different methylamines is
     initiated by the methyltransferases MtmB, MtbB, and MttB. A single
     in-frame amber (UAG) codon is found within each of the genes encoding
     these methylamine methyltransferases. Edman sequencing and   ***mass***
       ***spectroscopy***   of   ***purified***   MtmB   ***peptide***
```

fragments previously revealed that the UAG does not terminate translation, and that lysine is present at the amber encoded position. Recent x-ray crystallography and electrospray ***mass*** ***spectroscopy*** has shown that this lysyl residue is modified with a pyrroline ring in intact MtmB. This novel amino acid, apparently encoded by an amber codon, has been designated pyrrolysine. The pylT gene found near the mtmB genes in M. barkeri encodes a putative amber decoding tRNACUA. Northern analysis demonstrated that the tRNACUA gene is part of a larger 4.2 kb transcript. Both the 72 nt processed tRNACUA and 4.2 kb transcript are detectable during growth on either methanol or monomethylamine. One of the other genes on the 4.2 kb transcript, pylS, encodes a lysyl-tRNA synthetase (LysRS) only distantly related to other LysRS enzymes. Natural abundance tRNACUA was ***isolated*** from the total M. barkeri tRNA pool by hybridization to complementary oligonucleotides coupled to ***magnetic*** ***beads*** . A canonical lysine tRNA, tRNAUUU, was also ***isolated*** with similar methodology. Northern hybridizations indicated that these two ***purified*** tRNA species were not cross-contaminated. PylS was able to charge both tRNACUA and tRNAUUU with 14C-lysine. However, two other LysRS enzymes found in M. barkeri, LysK and LysS, were able to charge tRNAUUU, but not tRNACUA. It can be concluded that tRNACUA is specifically charged with lysine by PylS, one of the three LysRS enzymes found in M. barkeri. This appears to be the first step in the insertion of pyrrolysine at the UAG codon position. Current experimentation is aimed at determining whether the lys-tRNACUA serves as the substrate for the further synthesis of pyrrolysine (thus making this residue the 22nd translationally encoded amino acid) or if lysine is modified following its insertion into the ***protein*** at the UAG codon position.

L19 ANSWER 2 OF 2 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.
ACCESSION NUMBER: 2002:220586 BIOSIS
DOCUMENT NUMBER: PREV200200220586
TITLE: Homing and hematopoiesis: HCELL is the principal E-selectin and L-selectin ligand of human hematopoietic stem cells.
AUTHOR(S): Sackstein, Robert (1); Dimitroff, Charles J. (1); Lee, Jack Y. (1); Fuhlbrigge, Robert C. (1); Parmar, Kalindi; Mauch, Peter M.; Sandmaier, Brenda M.
CORPORATE SOURCE: (1) Dermatology and Medicine, Brigham and Women's Hospital, Boston, MA USA
SOURCE: Blood, (November 16, 2001) Vol. 98, No. 11 Part 1, pp. 710a. http://www.bloodjournal.org/. print.
Meeting Info.: 43rd Annual Meeting of the American Society of Hematology, Part 1 Orlando, Florida, USA December 07-11, 2001
ISSN: 0006-4971.
DOCUMENT TYPE: Conference
LANGUAGE: English

AB The selectins are becoming increasingly recognized for playing key roles in hematopoiesis. The endothelial selectins, E- and P-selectin, are both constitutively expressed on bone marrow (BM) microvascular endothelium, where they help mediate hematopoietic progenitor cell (HPC) migration into BM. Expression of the leukocyte selectin, L-selectin, on human CD34+ HPCs is associated with higher clonogenic activity in in vitro assays and faster engraftment following BM transplantation. Human HPCs also express PSGL-1, a ligand for all three selectins, however, paradoxically, engagement of PSGL-1 appears to inhibit clonogenic activity of human HPCs. These published data, collectively, have prompted us to explore the structure and distribution of selectin ligands expressed on human HPCs. Utilizing a new shear-based adhesion assay system developed in our laboratory, we have analyzed the cell surface glycoproteins of normal human HPCs that mediate L-, E- and P-selectin binding. Normal BM cells were separated into various lineage- and lineage+ subsets by ***magnetic*** ***bead*** sorting, and also sorted by flow cytometry of "side-population" cells following Hoechst dye staining. Cell membrane ***proteins*** were resolved into component bands by SDS-PAGE, then blotted onto PVDF. The blot was then placed in a flow chamber apparatus, and L-selectin+lymphocytes or stably transfected CHO cells bearing E- or P-selectin (designated CHO-E and CHO-P, respectively) were introduced into the chamber under controlled flow conditions. Adhesive interactions between cells in flow and immobilized (blot) ***proteins*** were visualized by video microscopy. CHO-P adhesive interactions occurred only at bands corresponding to PSGL-1. Adhesive interactions using lymphocytes

and CHO-E were also observed at bands corresponding to PSGL-1, but significantly more L- and E-selectin ligand activity was observed at a band of apprx100,000 mw, operationally named "Hematopoietic Cell E-/L-selectin Ligand" (HCELL). ***Mass*** ***spectroscopy*** analysis of this ***protein*** , confirmed by immunopurification, revealed that this E- and L-selectin ligand is a previously unrecognized glycoform of a well-characterized glycoprotein, CD44. In shear-based adhesion assays of ***purified*** ***protein*** or of ***protein*** expressed naturally on cell membranes, HCELL displays >5-fold more avidity for E- and for L-selectin compared to PSGL-1. Though CD44 is broadly expressed among normal human BM marrow cells, HCELL is expressed only on lineage- cells: its expression is characteristic of CD34+ cells, with highest expression in CD38-/lin- cells. Additionally, HCELL is expressed on CD34+ and CD34- subsets of "side-population" cells. The distinctive, restricted expression of HCELL among the subsets comprising the human hematopoietic "stem" cell and its marked avidity for E- and L-selectin supports a role for this unique glycoform of CD44 as a BM "homing receptor" as well as being the principal ligand mediating L-selectin-dependent cell-cell adhesive events within the BM microenvironment.

=> d his

(FILE 'HOME' ENTERED AT 13:48:38 ON 24 FEB 2003)

FILE 'MEDLINE, CAPLUS, BIOSIS, EMBASE, SCISEARCH, AGRICOLA' ENTERED AT 13:49:06 ON 24 FEB 2003

```
L1            1 S NANOMAG
L2      7884617 S PROTEIN OR PROTEINACEOUS OR PEPTIDE
L3      2053372 S SIOLAT? OR PURIF?
L4      5036901 S ISOLAT? OR PURIF?
L5      1223596 S L2 (P) L4
L6        15086 S (MAGNETIC PARTICLE) OR (MAGNETIC BEAD)
L7          847 S L5 (P) L6
L8         1856 S AGAROSE (P) HYDROPHOBIC
L9          937 S (SILICA GEL) (P) (REVERSE PHASE)
L10           0 S L7 (P) (L8 OR L9)
L11         231 S SILICA (P) MAGNETIC (P) (HYDROPHOBIC)
L12           0 S L5 (P) L11
L13      371943 S HYDROPHOBIC OR HYDROPHILIC
L14           7 S L7 (P) L13
L15           4 DUPLICATE REMOVE L14 (3 DUPLICATES REMOVED)
L16       60864 S MASS SPECTROSCOPY
L17           2 S L7 (P) L16
L18           2 DUPLICATE REMOVE L17 (0 DUPLICATES REMOVED)
L19           2 S L17 NOT L15
```

=> log y

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	73.24	73.45

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE ENTRY	TOTAL SESSION
CA SUBSCRIBER PRICE	-1.95	-1.95

STN INTERNATIONAL LOGOFF AT 13:59:05 ON 24 FEB 2003